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NEWS LETTER OF THE BUREAU OF ENTOMOLOGY.

U. S. DEPARTMENT OF AGRICULTURE.

Number 10. January-February, 1915.

NEW BOOKS IN LIBRARY.

Brun, Rudolf. Die Raumorientierung der Ameisen und das Orientierungsproblem im allgemeinen. Jena, Fischer, 1914. 234 p.

Das Tierreich. lfg. 41. Kieffer, J. J. Bethylidae, July, 1914.
lfg. 42. Kieffer, J. J. Serphidae. Calliceratidae. October, 1914.

Deutsche Entomologische Zeitschrift. Jahrgang 1911–1914, completing this set to date.

Froggatt, W. W. Pests and diseases of the coconut palm. (N. S. Wales Dept. Agr. Science Bulletin No. 2, ed. 3 rev. and enl., July, 1914.)

Hewitt, C. Gordon. The house-fly. Cambridge, at the University press, 1914. 382 p., illus., plates.
Bibliography: p. 336–372.

India. Dept. Agr. Memoirs, v. 4, no. 6 (Entomological series) July, 1914. Grove, A. J., and Ghosh, C. C. The life-history of Psylla isitis Buckt. (Psyllopa punctipennis, Crawford) The Psylla disease of indigo. p. 329–354, col. pl. XV–XX.

Johnson, J. W. Haigh. A contribution to the biology of sewage disposal, pt. 2. (Journal of economic biology, v. 9, no. 4, p. 127–164, illus. December, 1914.)
Bibliography: p. 1162–1164.

Minnesota State Entomologist. Fifteenth report . . . 1913 and 1914. (Ninth report of F. L. Washburn.) Agr. Exp. Sta., St. Anthony Park, Minn. December 1, 1914. 107 and 98 p. illus. col. pl.

Trägärdh, Ivar. Sveriges Skoginsekter [forest insects]. Stockholm, Hugo Gebers Förlag, 1914. 279 p illus. 16 pl. (Dr. Howard's personal copy available for consultation in Bureau Library.)

EDITORIAL WORK

The following publications have been issued since the December News Letter was mailed:

"Quassiin as a contact insecticide," Wm. B. Parker. Dept. Bull. 165, December 31, 1914.

"The chalcis fly in alfalfa seed," T. D. Urbahns. Farmer's Bull. 636, December 31, 1914.

"Susceptibility of citrus fruits to the attack of the Mediterranean fruit fly," E. A. Back and C. E. Pemberton. Jour. Agric. Research, Vol. III, No. 4, January 15, 1915.

"Three-cornered alfalfa hopper," V. L. Wildermuth. Jour. Agric. Research, Vol. III, No. 4, January 15, 191

"Life history of the Mediterranean fruit fly from the standpoint of parasite introduction," E. A. Back. Jour. Agric. Research, Vol. III, No. 5, February 15, 1915.

"Cactus solution as an adhesive in arsenical sprays for insects," M. M. High. Dept. Bull. 160, January 22, 1915.

"Wireworms attacking cereal and forage crops," J. A. Hyslop. Dept. Bull. 156, January 27, 1915.

"Para-dichlorobenzene as an insect fumigant," A. B. Duckett. Dept. Bull. 167, February 10, 1915.

"The European pine-shoot moth," August Busck. Dept. Bull. 170, February 9, 1915.

"A method for fumigating seed," E. R. Sasscer and Lon A. Hawkins. Dept. Bull. 186, February 27, 1915.

"Descriptions of some beetles reared from cotton in Peru," W. D. Pierce. Report 102, Office of the Secretary, January 25, 1915.

"The grasshopper problem and alfalfa culture," F. M. Webster. Farmer's Bull. 637, January 25, 1915.

"The Hessian fly," F. M. Webster. Farmer's Bull. 640, March 17, 1915.

"Alfalfa attacked by the clover-root curculio," F. M. Webster. Farmer's Bull. 649, February 27, 1915.

"Classification of the subfamily Cryphalinæ," A. D. Hopkins. Report 99, Office of the Secretary, March 10, 1915.

"Preliminary classification of the superfamily Scolytoidea," A. D. Hopkins. Technical Series Bull. 17, Part II, January 9, 1915. [Contents and index will follow.]

"The San Juan scale and its control," A. L. Quaintance. Farmer's Bull. 650, March 30, 1915.

"Cockroaches," C. L. Marlatt. Farmer's Bull. 658, March 27, 1915.

"The woolly apple aphis," A. C. Baker. Report 101, Office of the Secretary, March 31, 1915.

"Homemade lime-sulphur concentrate," E. W. Scott. Dept. Bull. 197, March 31, 1915.

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"Contents and index," Entomology Bull. 116, January 18, 1915.
"Contents and index," Entomology Bull. 115, February 5, 1915.
"Contents and index," Technical Series Bull. 27, February 13, 1915.
"Biology of the termites of the eastern United States," T. E. Snyder. Ent. Bull. 94, Part II, February 17, 1915.

THE STATUS OF ALL PART BULLETINS OF THE BUREAU OF ENTOMOLOGY PUBLISHED UNDER THE TITLE "BULLETINS, NEW SERIES," 1 TO 127, AND "TECHNICAL SERIES," 1 TO 27, ON APRIL 1, 1915.

Bulletin No. 58, Parts I–V, and Contents and Index, complete.
63, Parts I–VII, and Contents and Index, complete.
64, Parts I–X, and Contents and Index, complete.
66, Parts I–VII, and Contents and Index, complete.
68, Parts I–IX, and Contents and Index, complete.
75, Parts I–VII, and Contents and Index, complete.
80, Parts I–VIII, and Contents and Index, complete.
82, Parts I–VII, and Contents and Index, complete.
83, Part I. [Complete in 1 part. No index.]
85, Parts I–VIII, and Contents and Index, complete.
90, Parts I–III, and Contents and Index, complete.
94, Parts I–II, Contents and Index to follow.
95, Parts I–VII, and Contents and Index, complete.
96, Parts I–VI, Contents and Index to follow.
97, Parts I–VII, and Contents and Index, complete.
99, Parts I–II, Contents and Index to follow.
109, Parts I–VII, Contents and Index to follow.
115, Parts I–III, and Contents and Index, complete.
116, Parts I–V, and Contents and Index, complete.
127, Parts I–II, and Contents and Index, complete.

Technical Series:
Bulletin No. 12, Parts I–IX, and Contents and Index, complete.
16, Parts I–VII, Contents and Index to follow.
17, Parts I–II, Contents and Index to follow.
19, Parts I–V, Contents and Index to follow.
20, Parts I–VI, Contents and Index to follow.
23, Parts I–II, Contents and Index to follow.
25, Parts I–II, Contents and Index in press.
27, Parts I–II, and Contents and Index, complete.

Aside from the exceptions noted above, the publications of the Bureau of Entomology are complete in the following numbers:

Bulletins, new series, Nos. 1 to 127, inclusive.
Circulars, second series, Nos. 1 to 173, inclusive.
Technical Series Bulletins, Nos. 1 to 27, inclusive.

BEE CULTURE.

E. F. Phillips, *In Charge.*

Dr. E. F. Phillips attended the annual convention of the National Beekeepers' Association held in Denver, Colo., February 16–18.

Mr. E. R. Root, editor of Gleanings in Bee Culture, visited the Bee Culture Laboratory at Drummond, Md., on Monday, January 18.

Mr. W. T. Crossman has been given a temporary appointment at the Drummond laboratory.

Note.—Bulletin No. 114 of the Bureau of Entomology is embodied in Senate Document No. 305, entitled "Mexican Cotton-Boll Weevil."

CEREAL AND FORAGE INSECT INVESTIGATIONS.

F. M. Webster, *In Charge.*

The use of the label "bred from" in bureau notes was discontinued some time ago and only "reared from" labels have been used. Mr. G. G. Ainslie in a letter to Prof. Webster brings up the question whether "bred from" labels may not, after all, be of use with the especial significance that they apply only to material which has actually been "bred" in the sense of a breeder, that is the parent adults have been observed to copulate, the female to oviposit and the larva and pupa carried through to adult. This may apply also to the use of these terms in field notes. Discussion is invited.—L. O. H.

THE CELLOIDIN METHOD.

By Dr. Henry Fox.

It frequently happens that persons engaged in histological or embryological investigations find the ordinary paraffin method of embedding and sectioning unsuited to the material being studied. In such cases the use of the method here outlined may yield satisfactory results.

There are many modifications of the celloidin method, all of which are described in Lee's Microtomist's Vade-mecum, published by P. Blakiston's Son & Co., 1012 Walnut Street, Philadelphia. The particular method described here is that employed by the majority of workers. It is known as the wet method.

First, as to the advantages and disadvantages of the celloidin method as compared with the paraffin method it may be said that the only advantage found in the celloidin method is that it will frequently enable us to infiltrate objects which are either too large or too dense to be penetrated by paraffin and that the penetration of the celloidin can be effected without the use of heat. Paraffin on the other hand is suitable only for small and easily infiltrated objects, but where it is possible to use it, it should always be used for it is much more convenient and gives much finer results than the celloidin method. The latter has the following disadvantages: (1) Objects must remain in the celloidin bath a long time to secure adequate infiltration, the time required varying from one to three weeks or even longer depending on the size and density of the object; (2) the sections must be cut wet which necessitates the use of a horizontal knife and consequently the possession of a sliding microtome; (3) the sections do not adhere so that serial sections are obtained only with difficulty and by the use of special methods of mounting; (4) very thin sections can not be cut, it being very unusual to get any sections less than 0. 01 mm. in thickness and in the majority of cases it is necessary to cut them still thicker.

It should be borne in mind that celloidin and the substances used to dissolve it are very inflammable and should never be brought near a flame.

The celloidin used at the Charlottesville station was obtained from the Arthur H. Thomas Co., of Philadelphia. It comes in tightly stoppered glass bottles containing one ounce of the substance. The celloidin is in the form of shreds immersed in water.

To prepare solutions of the celloidin proceed as follows:

Remove several shreds of celloidin from the stock bottle and, if they have been kept immersed in water, dry them as thoroughly as possible with filter paper. Then weigh out 2½ grams and return any excess to the stock bottle. Wash the weighed celloidin shreds in 95 per cent alcohol for a few minutes, dry them on filter paper and allow them to remain exposed to the air in a warm place—say the top of the paraffin oven—for several hours. Take care to protect them from dust and do not under any circumstances bring them near a free flame. Allow the shreds to remain exposed to the air until they are thoroughly dry.

While the celloidin shreds are drying pour 25 c. c. absolute alcohol into an absolutely dry, wide-mouthed bottle. I use the shell vial, 10 cm. in height and 3 cm. in diameter, supplied by the bureau. Be sure that the bottle has a good, tight stopper. Add to the absolute alcohol placed in this bottle an equal volume of ether. Label the bottle "Absolute alcohol-ether mixture." Be sure that in measuring out these liquids the graduate used is perfectly dry so far as water is concerned. Keep the absolute alcohol-ether mixture tightly corked to prevent evaporation of the liquid, which is very volatile.

Add the 2½ grams of thoroughly dried celloidin to this absolute alcohol-ether mixture. Let this stand—shaking occasionally to hasten the solution—until the celloidin has dissolved and the whole has become a homogeneous, thick, sirupy mass. This may require a day,or two. Label the bottle "Thick celloidin."

Prepare a thin solution of celloidin as follows: Take another thoroughly dry, wide-mouthed bottle, similar to that already used, and pour into it about a third of the solution from the first or thick celloidin bottle. Add to this an approximately equal volume of a mixture of equal parts absolute alcohol and ether. Label the bottle "thin celloidin" and keep it tightly corked ready for use.

During the final stages of preparing the above solutions proceed to dehydrate the object to be embedded as follows:

(*a*) Assume the object to have been preserved in 70 per cent alcohol.

(*b*) Transfer it to 95 per cent alcohol, or, if the object is not easily handled, remove the 70 per cent alcohol with a pipette and replace it similarly with 95 per cent. The object should remain in this for a day as a general rule.

(*c*) Transfer the object to absolute alcohol. (In the case of objects not easily handled with a forceps, use the pipette as above, but be sure that the pipette used to transfer the absolute alcohol is perfectly dry.) Allow the object to stay in the absolute alcohol from 8 to 24 hours, depending on its size and penetrability.

(*d*) Transfer the object to a second bath of fresh absolute alcohol. This is to get rid of the last trace of any water that might possibly be in the object. Objects will not be satisfactorily infiltrated with the celloidin so long as any water remains in them. Where objects are not readily handled modify the transfer as indicated above.

(*e*) After from 8 to 24 hours remove the absolute alcohol with a *dry* pipette and replace it with a mixture of equal parts absolute alcohol and ether like that used in making up the celloidin solutions. Let the object stay in this mixture at least one day; it may remain in it indefinitely without harm.

Next transfer the object to the thin celloidin or else replace the absolute alcohol-ether mixture with it, using the precautions indicated above. Be sure that all instruments that come in contact with the liquids are perfectly dry. The object must remain in the thin celloidin for a considerable time, the exact duration of the bath depending on the character of the material. With most objects it is usually advisable to leave them in it a week.

Transfer the object as before to thick celloidin. It should remain in this about as long as in the thin celloidin.

When there is every reason to believe that the object has been thoroughly infiltrated with the celloidin, the next thing to do is to mount it. For this purpose we may proceed as follows:

(*a*) Take a water-color mold. Grease the inside of this with vaseline so that the entire inner surface is coated with a thin film. Fill the mold about one-fourth full of thick celloidin. Remove the object from the celloidin bath and place it in the mold where it will rest on the layer of celloidin and where it may be oriented into any desired position. Finally fill the mold with thick celloidin. In orienting the object in the celloidin use needles moistened with ether.

(*b*) When a film has formed over the exposed surface of the celloidin in the water-color mold transfer it carefully to a small flat-bottomed dish—a small stender dish, say—in which has been placed some chloroform to the depth of about 3 mm. The mold containing the celloidin need not be submerged in this. Keep the dish tightly covered to prevent evaporation of the chloroform. The celloidin mass should be left in here from one to three hours, but it will do no harm to leave it one or two days. The chloroform vapor hardens the celloidin, giving it the consistency of a tough jelly.

(*c*) When the celloidin has hardened sufficiently remove it from the mold. The film of vaseline usually enables one to readily detach it from the inner surface of the mold. The celloidin cube may then be placed back into the mold and the whole placed once more in chloroform vapor, where it should stay until ready to be mounted. Never let the celloidin dry out.

(*d*) Take a square block of wood of a size adapted to fit the holder of the microtome. Dip one end into ether and hold it there for a minute or two. Then dip the moistened end into thick celloidin. Try to do this quickly before the ether has had time to evaporate. This end will then be covered with a layer of celloidin. This should be allowed to dry until it becomes rather firm, so that it is not readily impressed with the finger. When this has taken place, take the cube of celloidin containing the embedded object and with a razor take a slice off the bottom so as to get a clean surface. Then quickly add a drop or two of thick celloidin to the celloidin-coated surface of the wooden block, moisten the cut end of the celloidin cube with ether and press it into the still soft celloidin which has just been added to the wooden block. Hold it there until the celloidin has become fairly firm and then place the block in a vessel to which a little chloroform has been added. This will complete the hardening in an hour or two.

The wooden block with the celloidin cube firmly cemented to it is now thrown into 70 per cent alcohol, where it may remain until the operator is ready to section the embedded object.

Before sectioning it is well to bear in mind that celloidin must never be allowed to dry. In my own practice I usually place two covered Syracuse watch glasses on the table next the microtome and in each of these I place 70 per cent alcohol. I also provide myself with a medium-size camel's-hair brush with which I transfer the alcohol from one of the dishes to the knife and to the celloidin and remove the sections from the knife to the second dish of alcohol.

In the work of sectioning proceed as follows:

(*a*) Arrange the microtome knife obliquely, so that it will slice through the object with a long drawing cut for at least half the length of the blade. (See fig. 32, p. 60, in Guyer's Animal Micrology, Univ. of Chicago Press, 1906.)

(*b*) Remove the mounted object from the alcohol and fasten the wooden block in the holder of the microtome. Then adjust this holder so that the celloidin is at the proper level for cutting.

(*c*) Throughout the operation keep the knife and the celloidin well flooded with 70 per cent alcohol.

(*d*) Draw the knife through the celloidin with a straight, steady stroke. Avoid pulling down or lifting the knife carrier.

(*e*) As each section is cut transfer it by means of a soft camel's-hair brush to a dish containing 70 per cent alcohol.

(*f*) If the microtome is not automatic, push the knife back to position before turning the screw which raises the celloidin block.

After the sections have been cut, the next step is to stain them. The practice here will vary somewhat according to the stain used. For ordinary work I prefer Delafield's hematoxylin.

Transfer the sections first to 50 per cent alcohol, 2 minutes.

Transfer to 35 per cent alcohol, 2 minutes.

Transfer to distilled water, 3 to 5 minutes.

Transfer to second bath of distilled water to get rid of all traces of alcohol, 5 minutes.

Transfer the sections to dilute Delafield's hematoxylin. I usually employ 1 part of the stock solution of hematoxylin to 9 parts distilled water. The duration of the staining will vary with different objects. It is well to remove, wash, and examine sections at intervals, and when the stain is found to be that desired the next step can be taken. By the use of the dilute stain overstaining is prevented. In the case of celloidin sections only the tissues of the sectioned object are stained; the celloidin matrix is not stained.

Wash sections in distilled water.

Transfer successively to 35, 50, and 70 per cent alcohols, leaving the sections from 2 to 3 minutes in each.

Transfer a few sections at a time to 95 per cent alcohol in which they may remain from 3 to 5 minutes.

Transfer them to carbol-xylol. This is a mixture of equal parts carbolic acid and xylol. In making it take a dry test tube, in it place enough crystals of *pure* carbolic acid to fill it about one-fourth of its length, melt these over a flame, allow the resulting liquid to cool, and then add an equal volume of xylol. Do not, however, bring the xylol near the flame. Each section should be allowed to remain in the carbol-xylol at least 10 minutes; a longer time will not injure them. The object of placing the sections in the carbol-xylol is to clear them.

(It will be observed that in the above process we pass directly from 95 per cent alcohol into our clearing fluid. This is because absolute alcohol dissolves celloidin and would thereby free the sections from the matrix which holds them together. Carbol-xylol is used because it clears objects which have not been completely dehydrated.)

Transfer the section to a slide; take up the excess of carbol-xylol by the use of a piece of filter paper, but do not let the section get entirely dry. Add a drop of Canada balsam, place a cover glass over it in the usual way and the mount is ready for study.

DECIDUOUS FRUIT INSECT INVESTIGATIONS.

A. L. Quaintance, *In Charge.*

Mr. B. R. Leach, who has been on furlough, attending Cornell University, since December 1, 1914, resumed his duties February 1, 1915.

In cooperation with Prof. C. P. Gillette, director and entomologist of the Colorado Agricultural Experiment Station, a laboratory has been established in the Grand Junction district of Colorado, where particular attention will be given to a study of the biology of the codling moth in that region. Extensive orchard spraying experiments are also planned. The codling moth in the Grand Valley has been for some years unusually destructive, and the methods of control successful in other regions do not here furnish satisfactory protection from its injuries.

Mr. E. H. Siegler, who has been attached to the Benton Harbor, Mich., laboratory, will be in immediate charge of the bureau work in the Grand Junction district.

The laboratory which has been in existence at Winthrop, Me., during the past two years will be discontinued, since the codling moth life-history studies in that region have now been completed and results prepared for publication.

Mr. F. L. Simanton will be transferred to the laboratory at Benton Harbor, Mich.

FOREST INSECT INVESTIGATIONS.

A. D. Hopkins, *In Charge.*

Messrs. H. G. Champion, of the University of Oxford and the Indian Imperial Forest Service, and T. E. Snyder spent some 10 days during January touring lumber mills in the high mountains of West Virginia in the study of the industry and the insects involved. While visiting the Union tannery at Davis, W. Va., they were informed by the manager that, following recommendations to prevent injury by the tanbark borer (*Dinoderus substriatus* Payk.), they used all hemlock tanbark before it was 4 years old. Dr. Hopkins visited this tannery in 1901 and found that out of a total of 20,000 cords of bark about 10,000 were badly damaged by this beetle. On close investigation, he found the damage was practically confined to bark that was from 2 to 7 years old, and no appreciable damage was done in bark less than 3 years old. Accordingly he recommended that, in order to avoid this loss, older bark be used first, and that no bark be kept for a longer period than 3 years. Thus, by following the suggestion then made, enormous loss has been prevented in this tannery alone.

SOUTHERN FIELD CROP INSECT INVESTIGATIONS.

W. D. Hunter, *In Charge.*

U. C. Loftin left for Cuba where he will be engaged for several months in the study and collection of parasites of sugar-cane insects and the investigation of the relation between certain systems of culture and the sugar-cane borer.

E. A. McGregor has arrived in Washington from his station at Batesburg, S. C., for consultations and studies which will keep him in the city for about a month.

D. L. Van Dine made a trip to Ithaca and New York City during January to confer with entomologists and members of the medical profession regarding his work on malarial mosquitoes.

W. V. King was in Washington for the purpose of obtaining data necessary to complete a thesis which he is soon to present to the faculty of Tulane University at New Orleans, La., for the degree of doctor of philosophy.

F. C. Bishopp, box 208, Dallas, Tex., has undertaken the taxonomic study of fleas. He has probably one of the largest collections in the United States at the Dallas laboratory but desires to obtain additional specimens from all parts of the country. It will assist greatly in his studies if material of this kind is sent directly to him.

TROPICAL AND SUBTROPICAL INSECT INVESTIGATIONS.

C. L. Marlatt, *In Charge.*

Mr. E. R. Sasscer, now with the Federal Horticultural Board, reports as a result of his inspection of the Introduction Garden at Miami the finding of a new and dangerous scale insect infesting mangoes, namely, *Lecanium* (*Coccus*) *mangiferae*. Incidentally he saw something of Mr. Yothers's work in Florida and reports as an eyewitness some remarkable demonstration results from spraying, in which the fruit output was enormously increased and the quality much improved as a result of the treatment.

Mr. J. R. Horton, in charge of the Louisiana citrus insect work, who was in Washington for consultation and work on his reports, has returned to New Orleans, La.

The Mediterranean fruit-fly force at Honolulu now includes, in addition to the leaders, Dr. Back and Mr. Pemberton, also Messrs. Willard, Banks, and Maxwell, Ah Fook, a Chinaman, and Muto, the Jap. Aside from Muto, these men are all connected with the inspection service.

The present arrangement puts Mr. Willard in general charge of this service under Dr. Back. Mr. Willard is a graduate of the Massachusetts Agricultural College, who came to the islands as instructor in agriculture and farm superintendent at the Mills Institute, Honolulu, and is reported by Dr. Back as being a very useful and serviceable assistant.

Dr. Back, with Mr. Pemberton, will be engaged during the next three months in an investigation of the fruit fly as affecting coffee, and other coffee insects of the Cona district, Island of Hawaii. This is the great coffee district of Hawaii. The pulp of the coffee berry is a favorite food of the Mediterranean fruit fly and frequently as many as a dozen maggots of this fly can be taken from a single coffee berry which is no larger than a medium-sized cherry. Of vast interest to the coffee industry and to the fruit-fly control on the island is the fact reported by Dr. Back that one of the parasites recently introduced on the island through the agency of the Territorial board of agriculture is already parasitizing the maggots in coffee berries to an average of upward of 50 per cent, in some instances reaching as high a percentage as 96. The parasite in question is *Opius humilis*. This seems to be another instance of the many successful parasite introductions which have been carried out on these islands.

WASHINGTON : GOVERNMENT PRINTING OFFICE : 1915